

SHORT COMMUNICATION

Chromosome Numbers for Two Species of *Damnacanthus* (Rubiaceae) from China

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Counts of the somatic chromosomes of two species of *Damnacanthus* from China are reported here for the first time: *Damnacanthus officinarum* was found to be diploid with $2n = 22+1B$, while *D. hainanensis* was found to be tetraploid with $2n = 44+1B$.

Key words: China, chromosome number, *Damnacanthus*

Damnacanthus C. F. Gaertn. (Rubiaceae) comprises 11 species of small evergreen shrubs, often occurring in natural laurel forests, of temperate to subtropical eastern Asia (Liao 1976, Lo 1979, Yamazaki 1987, 1993, Ruan 1999, Chen & Taylor 2011).

To date, chromosome counts have been published for nine species of *Damnacanthus* (Robrecht *et al.* 1991, Naiki & Nagamasu 2004). In the present study, we examined metaphase chromosomes in root tips of *Damnacanthus officinarum* C. C. Huang and *D. hainanensis* (H. S. Lo) H. S. Lo ex Y. Z. Ruan, two species not previ-

ously examined. Materials for *D. officinarum* were collected from three localities, and for *D. hainanensis* from one locality.

Materials and Methods

Living individuals *Damnacanthus officinarum* and *D. hainanensis* (Table 1) were brought to the laboratory of Nanchang University, Jiangxi, China. Several shoots with roots were cut from each individual and planted in soil; the remaining shoots were prepared to serve as voucher speci-

TABLE 1. Chromosome numbers, origin and voucher data for *Damnacanthus officinarum* C. C. Huang and *D. hainanensis* (H. S. Lo) H. S. Lo ex Y. Z. Ruan. N = number of individual(s) observed. All voucher specimens deposited at KYO.

Species	Locality	N	Count ($2n$)	Voucher
<i>D. officinarum</i>	1. Emei Shan, Sichuan, China	2	22+1B	<i>Li s.n.</i>
	2. Qingcheng Shan, Sichuan, China	1	22+1B	<i>Naiki and Wu 6061</i>
	3. Xingdou Shan, Hubei, China	1	22+1B	<i>Naiki et al. 6073</i>
<i>D. hainanensis</i>	4. Wuzhi Shan, Hainan, China	3	44+1B	<i>Naiki and Li 6065, 6066, 6067</i>

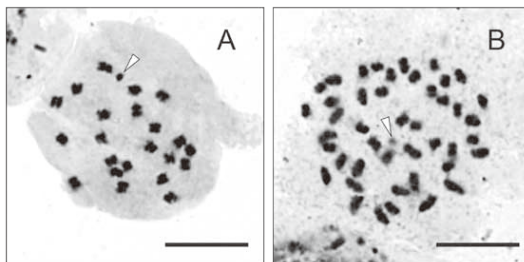


FIG. 1. Microphotographs of somatic chromosomes of *Damnacanthus*. A. *D. officinarum* (from Qingcheng Shan, locality 2 in Table 1), $2n = 22+1B$. B. *D. hainanensis* (from Wuzhi Shan, locality 4 in Table 1), $2n = 44+1B$. Arrow indicates satellite. Scale bar = $10\mu m$.

mens.

After about one month, freshly grown root tips were harvested and incubated in 0.002 M 8-hydroxyquinoline at $18^{\circ}C$ for 6 h, then fixed in Farmer's fixative (3 : 1 ethanol : glacial acetic acid) at $4^{\circ}C$ for at least 24 h. The root tips were macerated in 1 N hydrochloric acid at $60^{\circ}C$ for 5 min, then suspended in 45% acetic acid for at least 10 min. After hydrolysis, the root tip meristems were isolated and stained with 2% aceto-orcein for 30 min, then squashed. At least ten cells per individual were used to determine the number of chromosomes. The voucher specimens were deposited in the herbarium of Kyoto University (KYO).

Results and Discussion

We observed $2n = 22+1B$ somatic chromosomes in *Damnacanthus officinarum* and $2n = 44+1B$ in *D. hainanensis* (Table 1, Fig. 1). Because the basic chromosome number of *Damnacanthus* is $x = 11$ (Robbrecht *et al.*, 1991), we consider *D. officinarum* to be diploid and *D. hainanensis* to be tetraploid. The counts in the present study are the first for these two species.

Naiki & Nagamasu (2004) reported distyly in *Damnacanthus officinarum* based on an examination of herbarium specimens. Distyly is a genetically based dimorphism of the reproductive organs. Dimorphic plants comprise populations where flowers of some individuals have a long

style and short stamens, whereas other individuals have flowers with a short style and long stamens (Naiki 2012). Such dimorphism was not observed in 20 flowering individuals of *Damnacanthus hainanensis* in the present study. We were also unable to confirm distyly in *D. officinarum*, because we found only one or two individuals of *D. officinarum* at each locality cited in Table 1.

In *Damnacanthus*, correlation between distyly and ploidy level has been reported (Naiki & Nagamasu 2004). Diploid plants of *D.* showed distyly while tetraploid plants had monomorphic flowers in which the pistil is longer than the stamens. The results of our study coincide with those of Naiki (2012), who discussed the relationship between polyploidization and the breakdown of heterostyly.

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